

WHAT IS CLAIMED IS:

- 1 1. A single-stranded oligonucleotide molecule comprising or complementary
2 to a target sequence within a transcribed intronic RNA sequence of a target gene, wherein
3 the expression of said intronic RNA sequence has been determined to correlate with the
4 expression of an exonic mRNA sequence within said gene.
- 1 2. The single-stranded oligonucleotide molecule of claim 1 which is a PCR
2 primer or PCR probe.
- 1 3. The single-stranded oligonucleotide molecule of claim 2 which is a PCR
2 primer.
- 1 4. The single-stranded oligonucleotide molecule of claim 3 wherein said PCR
2 primer comprises or is complementary to a non-repetitive target sequence within said
3 transcribed intronic RNA sequence.
- 1 5. The single-stranded oligonucleotide molecule of claim 4 wherein said PCR
2 primer is a forward primer comprising 5'-sequences of said target sequence.
- 1 6. The single-stranded oligonucleotide molecule of claim 4 wherein said PCR
2 primer is a reverse primer complementing 5'-sequences of said target sequence.
- 1 7. The single-stranded oligonucleotide molecule of claim 3 wherein said
2 target sequence is at least 55 nucleotide bases long.
- 1 8. The single-stranded oligonucleotide molecule of claim 3 wherein said
2 target sequence is at least 60 nucleotide bases long.
- 1 9. The single-stranded oligonucleotide molecule of claim 3 wherein said PCR
2 primer is about 17-30 nucleotide bases in length.
- 1 10. The single-stranded oligonucleotide molecule of claim 9 wherein said PCR
2 primer contains about 20% to 80% G+C bases.

1 11. The single-stranded oligonucleotide molecule of claim 9 wherein said PCR
2 primer has a melting temperature (T_m) of between about 50 °C and about 70 °C.

1 12. The single-stranded oligonucleotide molecule of claim claim 2 which is a
2 PCR probe.

1 13. The single-stranded oligonucleotide molecule of claim 12 wherein said
2 PCR probe is labeled with a reporter fluorescent dye and a quencher fluorescent dye.

1 14. The single-stranded oligonucleotide molecule of claim 2 wherein the target
2 gene is selected from the group consisting of CEGP1, FOXM1, PRAME, and STK15.

1 15. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is CEGP1, and the PCR primer is selected from the group consisting of
3 forward and reverse primers of SEQ ID NOs: 14, 15, 17, 18, 20, 21, 23, and 24.

1 16. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is CEGP1, and the PCR probe is selected from the group consisting of SEQ
3 ID NOs: 16, 19, 22, and 25.

1 17. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is FOXM1, and the PCR primer is selected from the group consisting of
3 forward and reverse primers of SEQ ID NOs: 26, 27, 29, 30, 32, 33, 35, 36, 38, 39, 41,
4 42, 44, 45, 47, 48, 50, and 51.

1 18. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is FOXM1 and the PCR probe is selected from the group consisting of SEQ
3 ID NOs: 28, 31, 34, 37, 40, 43, 46, 49, and 52.

1 19. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is PRAME, and the PCR primer is selected from the group consisting of
3 forward and reverse primers of SEQ ID NOs: 53, 54, 56, and 57.

1 20. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is PRAME, and the PCR probe is selected from the group consisting of SEQ
3 ID NOs: 55 and 58.

1 21. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is STK15, and the PCR primer is selected from the group consisting of
3 forward and reverse primers of SEQ ID NOs: 59, 60, 62, 63, 65, and 66.

1 22. The single-stranded oligonucleotide molecule of claim 14 wherein the
2 target gene is STK15, and the PCR probe is selected from the group consisting of SEQ ID
3 NOs: 61, 64, and 67.

1 23. The single-stranded oligonucleotide molecule of claim 1 wherein the target
2 gene is selected from the group consisting of the genes listed in Figure 6.

1 24. The single-stranded oligonucleotide molecule of claim 1 wherein the target
2 gene is selected from the group consisting of β -actin; BAG1; bcl-2; CCNB1; CD68;
3 CEGP1; CTSL2; EstR1; GAPDH; GSTM1; GUS; GRB7; HER2; Ki-67; MYBL2; PR;
4 RPLPO; STK15; STMY3; SURVIVIN; and TFRC.

1 25. A method for monitoring gene expression in a biological sample,
2 comprising
3 (a) providing a polynucleotide complementary to an intronic RNA sequence
4 within a target gene, wherein the expression of said intronic RNA sequence correlates
5 with the expression of an exonic mRNA sequence within said gene;
6 (b) hybridizing said polynucleotide to said intronic RNA sequence to form a
7 polynucleotide-intronic RNA complex; and
8 (c) detecting the polynucleotide-intronic RNA complex.

1 26. The method of claim 25 wherein said intronic RNA sequence is selected
2 by identifying intronic sequences which are co-expressed with the mRNA of said target
3 gene, and selecting an intronic RNA sequence having the highest correlation coefficient
4 for said co-expression.

1 27. The method of claim 25 wherein said intronic RNA sequence is at least 50
2 nucleotide bases long.

1 28. The method of claim 25 wherein said biological sample is a tissue sample.

1 29. The method of claim 28 wherein said tissue is a tumor tissue.

1 30. The method of claim 29 wherein said tumor is cancer.

1 31. The method of claim 30 wherein said cancer is selected from the group
2 consisting of breast cancer, colon cancer, lung cancer, prostate cancer, hepatocellular
3 cancer, gastric cancer, pancreatic cancer, cervical cancer, ovarian cancer, liver cancer,
4 bladder cancer, cancer of the urinary tract, thyroid cancer, renal cancer, carcinoma,
5 melanoma, and brain cancer.

1 32. The method of claim 28 wherein said tissue sample is a fixed, wax-
2 embedded tissue sample.

1 33. The method of claim 32 wherein said exonic RNA is fragmented.

1 34. The method of claim 25 wherein said biological sample is a biological
2 fluid.

1 35. The method of claim 25 wherein said hybridization is performed under
2 stringent conditions.

1 36. The method of claim 25 further comprising the step of quantifying the
2 expression of said intronic RNA.

1 37. The method of claim 25 wherein said polynucleotide is a single-stranded
2 oligonucleotide.

1 38. The method of claim 37 wherein said single-stranded oligonucleotide is a
2 PCR probe or primer.

1 39. The method of claim 25 wherein the expression of more than one target
2 gene is monitored.

1 40. The method of claim 39 comprising simultaneous monitoring of a least 50
2 target genes.

1 41. The method of claim 39 comprising simultaneous monitoring of at least
2 500 target genes.

1 42. The method of claim 39 comprising simultaneous monitoring of at least
2 10,000 target genes.

1 43. The method of claim 42 wherein intronic RNA sequences corresponding to
2 a plurality of said target genes are displayed as an array immobilized on a solid surface.

1 44. The method of claim 25 wherein said target gene is selected from the
2 genes listed in Figure 6.

1 45. A method of preparing a single-stranded oligonucleotide molecule for
2 amplification of a target gene comprising:

3 (a) identifying at least one intron sequence within said target gene, the
4 expression of which correlates with the expression of an exonic mRNA sequence within
5 said target gene; and

6 (b) preparing a single-stranded oligonucleotide molecule that corresponds to at
7 least a portion of the transcribed intron sequence.

1 46. The method of claim 45 comprising identifying repeat sequences within
2 said intron sequence prior to preparing said single-stranded oligonucleotide molecule.

1 47. The method of claim 46 wherein said repeat sequences are masked prior to
2 preparing said oligonucleotide molecule.

1 48. The method of claim 45 wherein said single-stranded oligonucleotide
2 molecule is a PCR primer or probe.

1 49. The method of claim 48 wherein said PCR primer is a forward primer
2 designed to comprise 5'-sequences of a target sequence within said transcribed intron
3 sequence.

1 50. The method of claim 48 wherein said PCR primer is a reverse primer
2 designed to complement 5'-sequences of a target sequence downstream of the forward
3 primer within said transcribed intron sequence.

1 51. The method of claim 48 wherein said target sequence is at least 50
2 nucleotide bases long.

1 52. The method of claim 48 wherein said PCR probe is designed to comprise
2 or complement an internal portion of a target sequence within the transcribed intron
3 sequence.

1 53. The method of claim 52 wherein said PCR probe is labeled with a reporter
2 fluorescent dye and a quencher fluorescent dye.

1 54. The method of claim 48 wherein said target gene is selected from the
2 genes listed in Figure 6.

1 55. A method for amplifying intronic RNA in a fixed paraffin-embedded tissue
2 sample representing at least one gene of interest, comprising the steps of:

- 3 (a) contacting DNA obtained by reverse transcription of intronic RNA,
4 the expression of which correlates with the expression of a corresponding exonic RNA,
5 with at least one set of PCR primers and probe corresponding to said intronic RNA; and
6 (b) performing PCR amplification.

1 56. The method of claim 55 wherein said PCR primers and probe are designed
2 based upon a unique sequence within said intronic RNA.

1 57. The method of claim 56 wherein said sample comprises fragmented RNA
2 representing multiple genes of interest.

1 58. The method of claim 57 wherein said sample is contacted with a pool of
2 PCR primers and probes designed based upon unique sequences within introns, the
3 expression of which correlates with the expression of corresponding exons, present in
4 said genes of interest.

1 59. The method of claim 58 wherein said pool comprises at least one of the
2 intron-based primer/probe sets set forth in Figure 2.

1 60. The method of claim 58 wherein said pool comprises at least one forward
2 or reverse primer or probe set forth in Figure 2.

1 61. The method of claim 55 wherein said tissue sample is from a tumor biopsy.

1 62. The method of claim 61 wherein said tumor biopsy is obtained from a
2 human patient.

1 63. The method of claim 62 wherein said tumor is selected from the group
2 consistinh of breast cancer, lung cancer, and colorectal cancer.

1 64. The method of claim 55 further comprising the step of determining the
2 expression levels of the RNA transcripts of said genes of interest or their expression
3 products.

1 65. The method of claim 64 wherein differential expression of said RNA
2 transcripts or their products is correlated with predicted patient response to treatment or
3 patient survival.

1 66. The method of claim 55 wherein said gene of interest is selected from the
2 genes listed in Figure 6.

1 67. The method of claim 63 wherein the tumor is invasive breast cancer, and
2 the method comprises

3 (1) determining the expression levels of the RNA transcripts or
4 expression products of a gene or gene set selected from the group consisting of:

- 5 (a) Bcl2, cyclinG1, NFKBp65, NME1, EPHX1, TOP2B, DR5,
6 TERC, Src, DIABLO;
- 7 (b) Ki67, XIAP, hENT1, TS, CD9, p27, cyclinG1, pS2,
8 NFKBp65, CYP3A4;
- 9 (c) GSTM1, XIAP, Ki67, TS, cyclinG1, p27, CYP3A4, pS2,
10 NFKBp65, ErbB3;
- 11 (d) PR, NME1, XIAP, upa, cyclinG1, Contig51037, TERC,
12 EPHX1, ALDH1A3, CTSL;
- 13 (e) CA9, NME1, TERC, cyclinG1, EPHX1, DPYD, Src,
14 TOP2B, NFKBp65, VEGFC;
- 15 (f) TFRC, XIAP, Ki67, TS, cyclinG1, p27, CYP3A4, pS2,
16 ErbB3, NFKBp65;
- 17 (g) Bcl2, PRAME, cyclinG1, FOXM1, NFKBp65, TS, XIAP,
18 Ki67, CYP3A4, p27;
- 19 (h) FOXM1, cyclinG1, XIAP, Contig51037, PRAME, TS,
20 Ki67, PDGFRa, p27, NFKBp65;
- 21 (i) PRAME, FOXM1, cyclinG1, XIAP, Contig51037, TS, Ki6,
22 PDGFRa, p27, NFKBp65;
- 23 (j) Ki67, XIAP, PRAME, hENT1, contig51037, TS, CD9, p27,
24 ErbB3, cyclinG1;
- 25 (k) STK15, XIAP, PRAME, PLAUR, p27, CTSL, CD18,
26 PREP, p53, RPS6KB1;
- 27 (l) GSTM1, XIAP, PRAME, p27, Contig51037, ErbB3, GSTp,
28 EREG, ID1, PLAUR;
- 29 (m) PR, PRAME, NME1, XIAP, PLAUR, cyclinG1,
30 Contig51037, TERC, EPHX1, DR5;
- 31 (n) CA9, FOXM1, cyclinG1, XIAP, TS, Ki67, NFKBp65,
32 CYP3A4, GSTM3, p27;
- 33 (o) TFRC, XIAP, PRAME, p27, Contig51037, ErbB3, DPYD,
34 TERC, NME1, VEGFC; and

35 (p) CEGP1, PRAME, hENT1, XIAP, Contig51037, ErbB3,
 36 DPYD, NFKBp65, ID1, TS
 37 in said sample;
 38 (2) subjecting the data obtained in step (a) to statistical analysis; and
 39 (3) determining whether the likelihood of long-term survival of said
 40 patient, without the recurrence of breast cancer has increased or decreased.

1 68. The method of claim 67 wherein the expression levels of said RNA
 2 transcripts or their expression products are normalized against the expression levels of all
 3 RNA transcripts or their expression products in said breast cancer tissue sample, or of a
 4 reference set of RNA transcripts or their products.

1 69. The method of claim 63 wherein the tumor is estrogen receptor (ER)-
 2 positive invasive breast cancer, and the method comprises
 3 (1) determining the expression levels of the RNA transcripts or
 4 expression products of a gene or gene set selected from the group consisting of:
 5 (a) PRAME, p27, IGFBP2, HIF1A, TIMP2, ILT2, CYP3A4,
 6 ID1, EstR1, DIABLO;
 7 (b) Contig51037, EPHX1, Ki67, TIMP2, cyclinG1, DPYD,
 8 CYP3A4, TP, AIB1, CYP2C8;
 9 (c) Bcl2, hENT1, FOXM1, Contig51037, cyclinG1,
 10 Contig46653, PTEN, CYP3A4, TIMP2, AREG;
 11 (d) HIF1A, PRAME, p27, IGFBP2, TIMP2, ILT2, CYP3A4,
 12 ID1, EstR1, DIABLO;
 13 (e) IGF1R, PRAME, EPHX1, Contig51037, cyclinG1, Bcl2,
 14 NME1, PTEN, TBP, TIMP2;
 15 (f) FOXM1, Contig51037, VEGFC, TBP, HIF1A, DPYD,
 16 RAD51C, DCR3, cyclinG1, BAG1;
 17 (g) EPHX1, Contig51037, Ki67, TIMP2, cyclinG1, DPYD,
 18 CYP3A4, TP, AIB1, CYP2C8;
 19 (h) Ki67, VEGFC, VDR, GSTM3, p27, upa, ITGA7, rhoC,
 20 TERC, Pin1;
 21 (i) CDC25B, Contig51037, hENT1, Bcl2, HLAG, TERC,
 22 NME1, upa, ID1, CYP;

- 23 (j) VEGFC, Ki67, VDR, GSTM3, p27, upa, ITGA7, rhoC,
24 TERC, Pin1;
- 25 (k) CTSB, PRAME, p27, IGFBP2, EPHX1, CTSL, BAD, DR5,
26 DCR3, XIAP;
- 27 (l) DIABLO, Ki67, hENT1, TIMP2, ID1, p27, KRT19,
28 IGFBP2, TS, PDGFB;
- 29 (m) p27, PRAME, IGFBP2, HIF1A, TIMP2, ILT2, CYP3A4,
30 ID1, EstR1, DIABLO;
- 31 (n) CDH1; PRAME, VEGFC; HIF1A; DPYD, TIMP2,
32 CYP3A4, EstR1, RBP4, p27;
- 33 (o) IGFBP3, PRAME, p27, Bcl2, XIAP, EstR1, Ki67, TS, Src,
34 VEGF;
- 35 (p) GSTM3, PRAME, p27, IGFBP3, XIAP, FGF2, hENT1,
36 PTEN, EstR1, APC;
- 37 (q) hENT1, Bcl2, FOXM1, Contig51037, CyclinG1,
38 Contig46653, PTEN, CYP3A4, TIMP2, AREG;
- 39 (r) STK15, VEGFC, PRAME, p27, GCLC, hENT1, ID1,
40 TIMP2, EstR1, MCP1;
- 41 (s) NME1, PRAM, p27, IGFBP3, XIAP, PTEN, hENT1, Bcl2,
42 CYP3A4, HLAG;
- 43 (t) VDR, Bcl2, p27, hENT1, p53, PI3KC2A, EIF4E, TFRC,
44 MCM3, ID1;
- 45 (u) EIF4E, Contig51037, EPHX1, cyclinG1, Bcl2, DR5, TBP,
46 PTEN, NME1, HER2;
- 47 (v) CCNB1, PRAME, VEGFC, HIF1A, hENT1, GCLC,
48 TIMP2, ID1, p27, upa;
- 49 (w) ID1, PRAME, DIABLO, hENT1, p27, PDGFRa, NME1,
50 BIN1, BRCA1, TP;
- 51 (x). FBXO5, PRAME, IGFBP3, p27, GSTM3, hENT1, XIAP,
52 FGF2, TS, PTEN;
- 53 (y) GUS, HIA1A, VEGFC, GSTM3, DPYD, hENT1, EBXO5,
54 CA9, CYP, KRT18; and
- 55 (z) Bclx, Bcl2, hENT1, Contig51037, HLAG, CD9, ID1,
56 BRCA1, BIN1, HBEGF;

57 (2) subjecting the data obtained in step (1) to statistical analysis; and
58 (3) determining whether the likelihood of long-term survival of said
59 patient, without the recurrence of breast cancer has increased or decreased.

1 70. The method of claim 69 wherein the expression levels of said RNA
2 transcripts or their expression products are normalized against the expression levels of all
3 RNA transcripts or their expression products in said breast cancer tissue sample, or of a
4 reference set of RNA transcripts or their products.

1 71. The method of claim 63 wherein the tumor is breast cancer, and the
2 method comprises
3 (1) determining the expression levels of the RNA transcripts or
4 expression products of a gene or gene set selected from the group consisting of: FOXM1;
5 PRAME; SKT15, Ki-67; CA9; NME1; SURV; TFRC; YB-1; RPS6KB1; Src; Chk1;
6 CCNB1; Chk2; CDC25B; CYP3A4; EpCAM; VEGFC; hENT1; BRCA2; EGFR; TK1;
7 VDR; Bcl2; CEGP1; GSTM1; PR; BBC3; GATA3; DPYD; GSTM3; ID1; EstR1; p27;
8 XIAP; IGF1R; AK055699; P13KC2A; TGFB3; BAG1; pS2; WISP1; HNF3A; and
9 NFKBp65, normalized against the expression levels of all RNA transcripts or their
10 products in said sample, or of a reference set of RNA transcripts or their expression
11 products;
12 (2) subjecting the data obtained in step (a) to statistical analysis; and
13 (3) determining whether the likelihood of long-term survival of said
14 patient, without the recurrence of breast cancer has increased or decreased.

1 72. The method of claim 71 wherein the expression levels of said RNA
2 transcripts or their expression products are normalized against the expression levels of all
3 RNA transcripts or their expression products in said breast cancer tissue sample, or of a
4 reference set of RNA transcripts or their products.

1 73. The method of claim 63 wherein the tumor is invasive breast cancer, and
2 the method comprises determining the expression levels of RNA transcripts or expression
3 products of a gene or gene set from the group consisting of:
4 (a) p53BP2, Bcl2, BAD, EPHX1, PDGFR β , DIABLO, XIAP,
5 YB1, CA9, and KRT8;

- 6 (b) GRB7, CD68, TOP2A, Bcl2, DIABLO, CD3, ID1, PPM1D,
- 7 MCM6, and WISP1;
- 8 (c) PR, p53BP2, PRAME, DIABLO, CTSL, IGFBP2, TIMP1,
- 9 CA9, MMP9, and COX2;
- 10 (d) CD68, GRB7, TOP2A, Bcl2, DIABLO, CD3, ID1, PPM1D,
- 11 MCM6, and WISP1;
- 12 (e) Bcl2, p53BP2, BAD, EPHX1, PDGFR β , DIABLO, XIAP,
- 13 YB1, CA9, and KRT8;
- 14 (f) KRT14, KRT5, PRAME, p53BP2, GUS1, AIB1, MCM3,
- 15 CCNE1, MCM6, and ID1;
- 16 (g) PRAME, p53BP2, EstR1, DIABLO, CTSL, PPM1D,
- 17 GRB7, DAPK1, BBC3, and VEGFB;
- 18 (h) CTSL2, GRB7, TOP2A, CCNB1, Bcl2, DIABLO, PRAME,
- 19 EMS1, CA9, and EpCAM;
- 20 (i) EstR1, p53BP2, PRAME, DIABLO, CTSL, PPM1D,
- 21 GRB7, DAPK1, BBC3, and VEGFB;
- 22 (j) Chk1, PRAME, p53BP2, GRB7, CA9, CTSL, CCNB1,
- 23 TOP2A, tumor size, and IGFBP2;
- 24 (k) IGFBP2, GRB7, PRAME, DIABLO, CTSL, β -Catenin,
- 25 PPM1D, Chk1, WISP1, and LOT1;
- 26 (l) HER2, p53BP2, Bcl2, DIABLO, TIMP1, EPHX1, TOP2A,
- 27 TRAIL, CA9, and AREG;
- 28 (m) BAG1, p53BP2, PRAME, IL6, CCNB1, PAI1, AREG,
- 29 tumor size, CA9, and Ki67;
- 30 (n) CEGP1, p53BP2, PRAME, DIABLO, Bcl2, COX2,
- 31 CCNE1, STK15, and AKT2, and FGF18;
- 32 (o) STK15, p53BP2, PRAME, IL6, CCNE1, AKT2, DIABLO,
- 33 cMet, CCNE2, and COX2;
- 34 (p) KLK10, EstR1, p53BP2, PRAME, DIABLO, CTSL,
- 35 PPM1D, GRB7, DAPK1, and BBC3;
- 36 (q) AIB1, p53BP2, Bcl2, DIABLO, TIMP1, CD3, p53, CA9,
- 37 GRB7, and EPHX1
- 38 (r) BBC3, GRB7, CD68, PRAME, TOP2A, CCNB1, EPHX1,
- 39 CTSL GSTM1, and APC;

- (s) CD9, GRB7, CD68, TOP2A, Bcl2, CCNB1, CD3, DIABLO, ID1, and PPM1D;
- (t) EGFR, KRT14, GRB7, TOP2A, CCNB1, CTSL, Bcl2, TP, KLK10, and CA9;
- (u) HIF1 α , PR, DIABLO, PRAME, Chk1, AKT2, GRB7, CCNE1, TOP2A, and CCNB1;
- (v) MDM2, p53BP2, DIABLO, Bcl2, AIB1, TIMP1, CD3, p53, CA9, and HER2;
- (w) MYBL2, p53BP2, PRAME, IL6, Bcl2, DIABLO, CCNE1, EPHX1, TIMP1, and CA9;
- (x) p27, p53BP2, PRAME, DIABLO, Bcl2, COX2, CCNE1, STK15, AKT2, and ID1;
- (y) RAD51, GRB7, CD68, TOP2A, CIAP2, CCNB1, BAG1, IL6, FGFR1, and p53BP2;
- (z) SURV, GRB7, TOP2A, PRAME, CTSL, GSTM1, CCNB1, VDR, CA9; and CCNE2;
- (aa) TOP2B, p53BP2, DIABLO, Bcl2, TIMP1, AIB1, CA9, p53, KRT8, and BAD;
- (ab) ZNF217, GRB7, p53BP2, PRAME, DIABLO, Bcl2, COX2, CCNE1, APC4, and β -Catenin,
- in a breast cancer tissue sample obtained from said patient,
- (2) subjecting the data obtained in step (a) to statistical analysis; and
- (3) determining whether the likelihood of said long-term survival has increased or decreased.

74. The method of claim 73 wherein the expression levels of said RNA transcripts or their expression products are normalized against the expression levels of all RNA transcripts or their expression products in said breast cancer tissue sample, or of a reference set of RNA transcripts or their products.

75. A method for measuring gene expression using an array comprising a plurality of polynucleotides hybridizing to target genes of interest immobilized on a solid surface, wherein at least one of the said polynucleotides comprises an intron-based

4 sequence the expression of which correlates with the expression of a corresponding exon
5 sequence.

1 76. The method of claim 75 wherein all of said polynucleotides comprise
2 intron sequences.

1 77. The method of claim 75 comprising at least one of the amplicons shown in
2 Figures 1A-M, or the complement thereof.

1 78. The method of claim 75 comprising two or more of the amplicons shown
2 in Figures 1A-M, or the complement thereof.

1 79. The method of claim 75 comprising all of the amplicons shown in Figures
2 1A-M, or the complement thereof.

1 80. The method of claim 75 comprising using intron-based polynucleotide
2 sequences hybridizing to at least one gene of interest selected from the group consisting
3 of: FOXM1, PRAME, Bcl2, STK15, CEGP1, Ki-67, GSTM1, PR, BBC3, NME1, SURV,
4 GATA3, TFRC, YB-1, DPYD, CA9, Contig51037, RPS6K1 and Her2, wherein at least
5 80% of the sequences on said array are intron-based.

1 81. The method of claim 80 comprising using intron-based polynucleotide
2 sequences hybridizing to at least 5 of said genes.

1 82. The method of claim 80 comprising using intron-based polynucleotide
2 sequences hybridizing to at least 10 of said genes.

1 83. The method of claim 80 comprising using intron-based polynucleotide
2 sequences hybridizing to all of said genes.

1 84. The method of claim 75 comprising using intron-based polynucleotide
2 sequences hybridizing to at least one gene of interest selected from the group consisting
3 of: FOXM1, PRAME, Bcl2, STK15, CEGP1, Ki-67, GSTM1, CA9, PR, BBC3, NME1,
4 SURV, GATA3, TFRC, YB-1, DPYD, GSTM3, RPS6KB1, Src, Chk1, ID1, EstR1, p27,

5 CCNB1, XIAP, Chk2, CDC25B, IGF1R, AK055699, P13KC2A, TGFB3, BAG11,
6 CYP3A4, EpCAM, VEGFC, pS2, hENT1, WISP1, HNF3A, NFKBp65, BRCA2, EGFR,
7 TK1, VDR, Contig51037, pENT1, EPHX1, IF1A, CDH1, HIF1 α , IGFBP3, CTSB, Her2
8 and DIABLO.

1 85. The method of claim 84 comprising using intron-based polynucleotide
2 sequences hybridizing to at least 5 of said genes.

1 86. The method of claim 84 comprising using intron-based polynucleotide
2 sequences hybridizing to at least 10 of said genes.

1 87. The method of claim 84 comprising using intron-based polynucleotide
2 sequences hybridizing to all of said genes.

1 88. The method of claim 75 comprising using intron-based polynucleotide
2 sequences hybridizing to at least one gene set selected from the group consisting of:
3 (a) Bcl2, cyclinG1, NFKBp65, NME1, EPHX1, TOP2B, DR5,
4 TERC, Src, DIABLO;
5 (b) Ki67, XIAP, hENT1, TS, CD9, p27, cyclinG1, pS2,
6 NFKBp65, CYP3A4;
7 (c) GSTM1, XIAP, Ki67, TS, cyclinG1, p27, CYP3A4, pS2,
8 NFKBp65, ErbB3;
9 (d) PR, NME1, XIAP, upa, cyclinG1, Contig51037, TERC,
10 EPHX1, ALDH1A3, CTSL;
11 (e) CA9, NME1, TERC, cyclinG1, EPHX1, DPYD, Src,
12 TOP2B, NFKBp65, VEGFC;
13 (f) TFRC, XIAP, Ki67, TS, cyclinG1, p27, CYP3A4, pS2,
14 ErbB3, NFKBp65;
15 (g) Bcl2, PRAME, cyclinG1, FOXM1, NFKBp65, TS, XIAP,
16 Ki67, CYP3A4, p27;
17 (h) FOXM1, cyclinG1, XIAP, Contig51037, PRAME, TS,
18 Ki67, PDGFR α , p27, NFKBp65;
19 (i) PRAME, FOXM1, cyclinG1, XIAP, Contig51037, TS, Ki6,

- 20 PDGFRa, p27, NFKBp65;
- 21 (j) Ki67, XIAP, PRAME, hENT1, contig51037, TS, CD9, p27,
- 22 ErbB3, cyclinG1;
- 23 (k) STK15, XIAP, PRAME, PLAUR, p27, CTSL, CD18,
- 24 PREP, p53, RPS6KB1;
- 25 (l) GSTM1, XIAP, PRAME, p27, Contig51037, ErbB3, GSTp,
- 26 EREG, ID1, PLAUR;
- 27 (m) PR, PRAME, NME1, XIAP, PLAUR, cyclinG1,
- 28 Contig51037, TERC, EPHX1, DR5;
- 29 (n) CA9, FOXM1, cyclinG1, XIAP, TS, Ki67, NFKBp65,
- 30 CYP3A4, GSTM3, p27;
- 31 (o) TFRC, XIAP, PRAME, p27, Contig51037, ErbB3, DPYD,
- 32 TERC, NME1, VEGFC; and
- 33 (p) CEGP1, PRAME, hENT1, XIAP, Contig51037, ErbB3,
- 34 DPYD, NFKBp65, ID1, TS.

- 1 89. The method of claim 75 comprising using intron-based polynucleotide
- 2 sequences hybridizing to at least one gene set selected from the group consisting of:
- 3 (a) PRAME, p27, IGFBP2, HIF1A, TIMP2, ILT2, CYP3A4,
- 4 ID1, EstR1, DIABLO;
- 5 (b) Contig51037, EPHX1, Ki67, TIMP2, cyclinG1, DPYD,
- 6 CYP3A4, TP, AIB1, CYP2C8;
- 7 (c) Bcl2, hENT1, FOXM1, Contig51037, cyclinG1,
- 8 Contig46653, PTEN, CYP3A4, TIMP2, AREG;
- 9 (d) HIF1A, PRAME, p27, IGFBP2, TIMP2, ILT2, CYP3A4,
- 10 ID1, EstR1, DIABLO;
- 11 (e) IGF1R, PRAME, EPHX1, Contig51037, cyclinG1, Bcl2,
- 12 NME1, PTEN, TBP, TIMP2;
- 13 (f) FOXM1, Contig51037, VEGFC, TBP, HIF1A, DPYD,
- 14 RAD51C, DCR3, cyclinG1, BAG1;
- 15 (g) EPHX1, Contig51037, Ki67, TIMP2, cyclinG1, DPYD,
- 16 CYP3A4, TP, AIB1, CYP2C8;
- 17 (h) Ki67, VEGFC, VDR, GSTM3, p27, upa, ITGA7, rhoC,
- 18 TERC, Pin1;

- 19 (i) CDC25B, Contig51037, hENT1, Bcl2, HLAG, TERC,
20 NME1, upa, ID1, CYP;
- 21 (j) VEGFC, Ki67, VDR, GSTM3, p27, upa, ITGA7, rhoC,
22 TERC, Pin1;
- 23 (k) CTSB, PRAME, p27, IGFBP2, EPHX1, CTSL, BAD, DR5,
24 DCR3, XIAP;
- 25 (l) DIABLO, Ki67, hENT1, TIMP2, ID1, p27, KRT19,
26 IGFBP2, TS, PDGFB;
- 27 (m) p27, PRAME, IGFBP2, HIF1A, TIMP2, ILT2, CYP3A4,
28 ID1, EstR1, DIABLO;
- 29 (n) CDH1; PRAME, VEGFC; HIF1A; DPYD, TIMP2,
30 CYP3A4, EstR1, RBP4, p27;
- 31 (o) IGFBP3, PRAME, p27, Bcl2, XIAP, EstR1, Ki67; TS, Src,
32 VEGF;
- 33 (p) GSTM3, PRAME, p27, IGFBP3, XIAP, FGF2, hENT1,
34 PTEN, EstR1, APC;
- 35 (q) hENT1, Bcl2, FOXM1, Contig51037, CyclinG1,
36 Contig46653, PTEN, CYP3A4, TIMP2, AREG;
- 37 (r) STK15, VEGFC, PRAME, p27, GCLC, hENT1, ID1,
38 TIMP2, EstR1, MCP1;
- 39 (s) NME1, PRAM, p27, IGFBP3, XIAP, PTEN, hENT1, Bcl2,
40 CYP3A4, HLAG;
- 41 (t) VDR, Bcl2, p27, hENT1, p53, PI3KC2A, EIF4E, TFRC,
42 MCM3, ID1;
- 43 (u) EIF4E, Contig51037, EPHX1, cyclinG1, Bcl2, DR5, TBP,
44 PTEN, NME1, HER2;
- 45 (v) CCNB1, PRAME, VEGFC, HIF1A, hENT1, GCLC,
46 TIMP2, ID1, p27, upa;
- 47 (w) ID1, PRAME, DIABLO, hENT1, p27, PDGFRa, NME1,
48 BIN1, BRCA1, TP;
- 49 (x) FBXO5, PRAME, IGFBP3, p27, GSTM3, hENT1, XIAP,
50 FGF2, TS, PTEN;
- 51 (y) GUS, HIA1A, VEGFC, GSTM3, DPYD, hENT1, EBXO5,
52 CA9, CYP, KRT18; and

53 (z) Bclx, Bcl2, hENT1, Contig51037, HLAG, CD9, ID1,
54 BRCA1, BIN1, HBEGF.

1 90. The method of claim 75 comprising using intron-based polynucleotide
2 sequences hybridizing to at least one gene set selected from the group consisting of:

- 3 (a) p53BP2, Bcl2, BAD, EPHX1, PDGFR β , DIABLO, XIAP,
4 YB1, CA9, and KRT8;
5 (b) GRB7, CD68, TOP2A, Bcl2, DIABLO, CD3, ID1,
6 PPM1D,
7 MCM6, and WISP1;
8 (c) PR, p53BP2, PRAME, DIABLO, CTSL, IGFBP2, TIMP1,
9 CA9, MMP9, and COX2;
10 (d) CD68, GRB7, TOP2A, Bcl2, DIABLO, CD3, ID1,
11 PPM1D,
12 MCM6, and WISP1;
13 (e) Bcl2, p53BP2, BAD, EPHX1, PDGFR β , DIABLO, XIAP,
14 YB1, CA9, and KRT8;
15 (f) KRT14, KRT5, PRAME, p53BP2, GUS1, AIB1, MCM3,
16 CCNE1, MCM6, and ID1;
17 (g) PRAME, p53BP2, EstR1, DIABLO, CTSL, PPM1D,
18 GRB7, DAPK1, BBC3, and VEGFB;
19 (h) CTSL2, GRB7, TOP2A, CCNB1, Bcl2, DIABLO,
20 PRAME,
21 EMS1, CA9, and EpCAM;
22 (i) EstR1, p53BP2, PRAME, DIABLO, CTSL, PPM1D,
23 GRB7, DAPK1, BBC3, and VEGFB;
24 (j) Chk1, PRAME, p53BP2, GRB7, CA9, CTSL, CCNB1,
25 TOP2A, tumor size, and IGFBP2;
26 (k) IGFBP2, GRB7, PRAME, DIABLO, CTSL, β -Catenin,
27 PPM1D, Chk1, WISP1, and LOT1;
28 (l) HER2, p53BP2, Bcl2, DIABLO, TIMP1, EPHX1, TOP2A,
29 TRAIL, CA9, and AREG;
30 (m) BAG1, p53BP2, PRAME, IL6, CCNB1, PAI1, AREG,
31 tumor size, CA9, and Ki67;

- 32 (n) CEGP1, p53BP2, PRAME, DIABLO, Bcl2, COX2,
- 33 CCNE1, STK15, and AKT2, and FGF18;
- 34 (o) STK15, p53BP2, PRAME, IL6, CCNE1, AKT2, DIABLO,
- 35 cMet, CCNE2, and COX2;
- 36 (p) KLK10, EstR1, p53BP2, PRAME, DIABLO, CTSL,
- 37 PPM1D, GRB7, DAPK1, and BBC3;
- 38 (q) AIB1, p53BP2, Bcl2, DIABLO, TIMP1, CD3, p53, CA9,
- 39 GRB7, and EPHX1
- 40 (r) BBC3, GRB7, CD68, PRAME, TOP2A, CCNB1, EPHX1,
- 41 CTSL GSTM1, and APC;
- 42 (s) CD9, GRB7, CD68, TOP2A, Bcl2, CCNB1, CD3,
- 43 DIABLO, ID1, and PPM1D;
- 44 (t) EGFR, KRT14, GRB7, TOP2A, CCNB1, CTSL, Bcl2, TP,
- 45 KLK10, and CA9;
- 46 (u) HIF1 α , PR, DIABLO, PRAME, Chk1, AKT2, GRB7,
- 47 CCNE1, TOP2A, and CCNB1;
- 48 (v) MDM2, p53BP2, DIABLO, Bcl2, AIB1, TIMP1, CD3,
- 49 p53, CA9, and HER2;
- 50 (w) MYBL2, p53BP2, PRAME, IL6, Bcl2, DIABLO, CCNE1,
- 51 EPHX1, TIMP1, and CA9;
- 52 (x) p27, p53BP2, PRAME, DIABLO, Bcl2, COX2, CCNE1,
- 53 STK15, AKT2, and ID1;
- 54 (y) RAD51, GRB7, CD68, TOP2A, CIAP2, CCNB1, BAG1,
- 55 IL6, FGFR1, and p53BP2;
- 56 (z) SURV, GRB7, TOP2A, PRAME, CTSL, GSTM1, CCNB1,
- 57 VDR, CA9; and CCNE2;
- 58 (aa) TOP2B, p53BP2, DIABLO, Bcl2, TIMP1, AIB1, CA9,
- 59 p53, KRT8, and BAD; and
- 60 (ab) ZNF217, GRB7, p53BP2, PRAME, DIABLO, Bcl2,
- 61 COX2, CCNE1, APC4, and β -Catenin.

1 91. The method of claim 75 comprising intron-based polynucleotide sequences
2 hybridizing to at least one genes selected from the group consisting of: CD68; CTSL;
3 FBXO5; SURV; CCNB1; MCM2; Chk1; MYBL2; HIF1A; cMET; EGFR; TS; STK15,

4 IGFR1; BCL2; HNF3A; TP53BP2; GATA3; BBC3; RAD51C; BAG1; IGFBP2; PR;
5 CD9; RB1; EPHX1; CEGP1; TRAIL; DR5; p27; p53; MTA; RIZ1; ErbB3; TOP2B;
6 EIF4E, CD68; CTSL; FBXO5; SURV; CCNB1; MCM2; Chk1; MYBL2; HIF1A; cMET;
7 EGFR; TS; and STK15.

1 92. The method of claim 75 comprising intron-based polynucleotide sequences
2 hybridizing to at least one genes selected from the group consisting of B-actin; BAG1;
3 bcl-2; CCNB1; CD68; CEGP1; CTSL2; EstR1; GAPDH; GSTM1; GUS; GRB7; HER2;
4 Ki-67; MYBL2; PR; RPLPO; STK15; STMY3; SURVIVIN; and TFRC.

1 93. The method of claim 75 comprising using intron-based polynucleotide
2 sequences corresponding to at least one gene selected from the genes listed in Figure 6.

1 94. The method of claim 75 comprising using intron-based polynucleotide
2 sequences corresponding to a plurality of genes selected from the genes listed in Figure 6.

1 95. The method of claim 75 comprising using both intron-based and exon-
2 based polynucleotide sequences.

1 96. The method of claim 95 comprising using both intron-based and exon-
2 based polynucleotide sequences hybridizing to the same target gene of interest.

1 97. The method of claim 75 wherein said array comprises at least 100 genes.

1 98. The method of claim 75 wherein said array comprising at least 100 genes
2 in a 100 μ section.

1 99. The method of claim 75 wherein said array comprises at lest 150 genes in a
2 100 μ section.